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Evaluation/Refinement of Fall Armyworm, Spodoptera Frugiperda (J.E. Smith), Thresholds, in Mississippi Whorl Stage Field Corn And Grain Sorghum

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By

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During 2016 and 2017, studies were conducted to determine the effects of *Spodoptera frugiperda* (J.E. Smith) on both damage and yield of *Sorghum bicolor* (L.) Moench and *Zea mays* (L.) when infested during the whorl stage. Results from damage ratings suggest that as the amount of overall plants infested increased, overall damage rating increased. However, yield results suggested that there was no yield loss as percent infested plants increased. Other studies were conducted to determine the most sensitive vegetative growth stages of grain sorghum and field corn. Manual damage studies suggests that extensive damage to field corn during growth stages V9 to V15 will cause significant yield loss. Also, damage to grain sorghum after growth stage V8 and prior to boot stage can cause significant yield loss.

DEDICATION

I would first like to dedicate this research to my beautiful wife, Rachel, for her constant patience and love throughout this entire process. This degree was completed in an effort to provide a better life for us in the near future. None of this would be possible without your support and encouragement.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

LITERATURE REVIEW

Field Corn

The most commonly and widely grown feed grain in the United States is field corn, *Zea mays* (L.), with an overall value of $51,700,000 in 2016 (USDA 2017). Approximately 303,500 ha (750,000 acres) of all-purpose corn was planted in Mississippi during 2016. There were 291,400 ha (720,000 acres) harvested strictly for grain at an average yield of 10,419 kg/ha (166 bushels per acre), and 4,000 ha (10,000 acres) were harvested for silage with an average yield of 31.38 megagrams per hectare (14 tons per acre). The overall value of Mississippi production was $448,200,000 for 2016 and was second only to soybean in value (NASS 2017).

Zea is the genus which belongs to the grass family of Poaceae (Doebley 1990). The origin for *mays* has been identified as the Mesoamerican region, now more commonly known as Mexico and Central America (Watson & Dallwitz 1992). Corn is a monoecious plant that is self and cross-pollinated. The male portion of maize is known as the tassel or staminate. The female portion is the pistillate inflorescence or the female flower (Purseglove 1972). Pollination will occur as pollen is transferred from the tassel to the silk of the ear as it falls. During pollination, 95% of the ovules are cross-pollinated from nearby plants and 5% are self-pollinated (Poehlman 1959). There are several different ways to describe the growth stages of a corn plant.

Corn growth and development can be separated into two periods, vegetative and reproductive. The growth stages within these periods are described in Abendroth et al. (2011). The vegetative period consists of growth stages from seedling emergence (VE) to tassel emergence (VT). The growth stages following seedling emergence (VE) up to tassel emergence (VT) are described based on the number of leaf collars present (V1-Vn). Reproductive growth stages are described based on ear development events, beginning at silk emergence (R1) and continuing until physiological maturity, i.e. black layer (R6), and include blister (R2), milk (R3), dough (R4), and dent (R5). Basic characteristics of each reproductive stage include (R1): silks become visible outside the husk, (R2): kernels begin to turn white, (R3): fluid inside begins to turn to a milky white, (R4): the fluid starts to turn pasty, (R5): all kernels begin to dent at the top, (R6): all endosperm within the kernel has dried down and black layer has been reached (Hanway and Ritchie 1997).

Although there are a range of relative maturities for commercial corn hybrids, corn growth and development is directly influenced by temperature. Development can be described using Growing Degree Units/Day, with a critical GDU threshold for each growth stage. Growing Degree Units are calculated using the formula (GDU=[((Tmax+Tmin)/2)-Tbase], where Tmax is maximum daily temperature, Tmin is minimum daily temperature, and Tbase is minimum temperature for corn growth and development (10°C) (Stewart et al. 1998, Dwyer et al. 1999). For the state of Mississippi the optimum planting dates range from mid-February to late April. Most relative maturity corn hybrids planted in Mississippi range from 110 to 120 day relative maturity (Larson 2012). Earlier planted corn often avoids infestations of some insect pests.

There are numerous insect pests that can effect corn in the Mid-South region of the United States. Early season corn pests can have a very detrimental effect by damaging the seeds and emerged seedlings. Corn seed and seedlings can be greatly affected by below ground insect pests including seedcorn maggot, *Delia platura* (Meigen); white grubs, *Phyllophaga spp*; Japanese beetle, *Popillia japonica* (Newman); southern corn rootworm, *Diabrotica undecimpunctata howardi* (Barber); wireworms, *Melanotus spp*.; lesser cornstalk borer, *Elasmopalpus lignosellus* (Zeller); corn leaf aphid, *Rhopalosiphum maidis* (F.); billbugs, *Sphenophorus spp*; chinch bug, *Blissus leucopterus leucopterus* (Say); black cutworm, *Agrotis ipsilon* (Hufnagel); sugarcane beetle, *Euetheola humilis rugiceps* (LeConte); brown stink bug, *Euschistus servus* (Say); green stink bug, *Acrosternum hilare* (Say); and southern green stink bug, *Nezara viridula* (L.) (Steffey et al. 1999, Akin et al. 2012, Catchot et al. 2018). During the vegetative stage: corn earworm, *Helicoverpa zea* (Boddie); true armyworm, *Pseudaletia unipuncta* (Haworth); fall armyworm, *Spodoptera frugiperda* (J.E. Smith); European corn borer, *Ostrinia nubilalis* (Hübner); Southwestern corn borer, *Diatraea grandiosella* (Dyar); stink bugs and grasshoppers can infest plants (Steffey et al. 1999, Akin et al. 2012, Catchot et al. 2018). Corn borers, corn earworm, fall armyworm, Japanese bettle, *Moodna bisinuella* (Hampson), and stink bugs can injure corn plants during the reproductive stages from pre-tassel and to the black layer stage (Steffey et al. 1999, Akin et al. 2012, Catchot et al. 2018).

Grain Sorghum

Grain sorghum, *Sorghum bicolor* (L.) Moench, is one of the top five cereal crops that is grown throughout the world. The United States is the largest producer with over 12 billion kilograms produced at an average yield of 5,238.86 kilograms per hectare (77.9 bushels per acre) (USCP 2016). Grain sorghum is the third-largest cereal grain grown in the United States based on scale of production. It is mainly grown in the south central United States from southern Nebraska and to the southern portions of Texas. In Mississippi, 3,600 ha of sorghum were harvested in 2017 down from 4,400 ha in 2016 (NASS 2017). Sorghum is well suited for this area because of its drought tolerance, resistance to mycotoxins and fungi, and its survivability in harsh climates (U.S. Grains Council 2017). A common use of grain sorghum in both the United States and across the globe is in livestock feed (Nuessly et al. 2013). In 2016, 21% of all sorghum produced in the United States was used for ethanol production followed by 15% for livestock feed. All remaining sorghum produced in the United States is exported to other countries (USCP 2016). The leading importers were China, Mexico, and Pakistan with China receiving over 7 billion kilograms (USCP 2016).

The optimum soil temperature for sorghum emergence and development is 21°C, but emergence will begin at a minimum temperature of 12.5°C (Stichler et al. 1997). In Mississippi, planting dates range from mid-April to mid-May. The seeding rate of grain sorghum can vary significantly based on the geography of the area being planted. The range could be from 49,000 to 270,000 plants per hectare with a row spacing of 91-101 cm (Stichler et al. 1997). Grain sorghum is known to be a drought tolerant crop that can be planted in arid regions of the United States (Stichler et al. 1997). The growth stages of grain sorghum are identified as stages 0-9. The vegetative growth stages 0-2 are described as (0) emergence, (1) early vegetative growth, (2) late vegetative growth (Vanderlip 1993). Reproductive growth stages consist of stages 3-9 and are described as

(3) early development of reproductive structures, (4) flag leaf emergence, (5) boot stage, (6) half bloom, during which grain formation begins, (7) soft dough stage, (8) hard dough stage, (9) physiological maturity (Vanderlip 1993, Gerik et al. 2003). The overall time between flowering and physiological maturity will vary depending on hybrid and environmental conditions. This entire process from flowering to physiological maturity will last one-third of the entire growing cycle (Vanderlip 1993).

Grain sorghum has a variety of pests that can affect production throughout the season. The most economically important pests of grain sorghum in the southern portion of the United States are lesser cornstalk borer; black cutworm; chinch bug; fire ant, *Solenopsis invicta* (Buren); sorghum midge, *Stenodiplosis sorghicola* (Coquillett); sorghum webworm, *Nola sorghiella* (Riley); fall armyworm; corn earworm; and the sugarcane aphid, *Melanaphis sacchari* (Zehntner), which was discovered in Louisiana, Texas, and Mississippi in 2013 (Cronholm et al. 1998, Elliott et al. 2015).

Fall armyworm can be both a direct and indirect pest depending on when infestations occur. Fall armyworm larvae will feed in the whorl of sorghum plants and cause indirect damage. They can also cause direct damage by feeding on developing grain and causing immediate yield loss. Planting dates can be manipulated to reduce the risk of large infestations of fall armyworm and other insect pests.

Fall Armyworm

The fall armyworm is of tropical origin and can be found overwintering in the United States in areas of southern Florida and southern Texas. It has been known to survive in certain years along the Gulf Coast (Lunginbill 1928). The fall armyworm has no diapause mechanism and will overwinter in warm regions where host plants are available (Sparks 1979). It is known to feed on more than 80 species of plants. The most common found in the southeast are crabgrass, *Digitaria sanguinalis* (L.) Scop.; cotton, *Gossypium hirsutum* (L.); corn, *Zea mays* (L.), sweet corn, *Zea mays* (L.) *var. saccharata;* soybean, *Glycine max* (L.) Merr.; sorghum, *Sorghum bicolor* (L.) Moench; morning glory, *Ipomoea purpurea* (L.) Roth; and Bermudagrass, *Cynodon dactylon* (L.) Pers. (Lunginbill 1928).

The fall armyworm adult is known to be nocturnal. During this time, adults will mate, oviposit, and feed. Following the feeding period virgin females will begin calling (Sparks 1979). Males can respond to the female from up to 40 feet (Sparks 1979). Fall armyworm females commonly oviposit on the underside of lower leaves, with the eggs laid in clusters of a few to hundreds of eggs protected by a covering of scales. Eggs will hatch in 2-4 days and neonates begin feeding on foliage. Larvae will complete six instars before pupation (Sparks 1979). During the first through third instar stages, larvae consume $< 2\%$ of the total amount of foliage that will be consumed during the larval stages. The remainder (98%) is consumed during the third through sixth instar (Sparks 1979). Pupation occurs in the soil, and this stage can last 7 to 37 days depending on soil temperature. The entire lifecycle of the fall armyworm ranges from 30 to 90 days. In warmer areas, such as the southeastern U.S. and around the Gulf Coast states, the complete lifecycle is 30 days during the summer (Vickery 1929).

The fall armyworm is an economically important pest in the southern United States. It is known to occur every year in the southern region and will feed on and damage an array of crops (Luginbill 1928). Major outbreaks in the United States were recorded during 1899 and 1912 in several crops including corn, grain sorghum, rice, vegetables, and cotton. In some areas of the South, this resulted in corn having to be replanted up to four times, and severe losses in sorghum (Luginbill 1928). According to Sparks (1979), during the years of 1975-1977 there were large infestations of the fall armyworm along the Atlantic coast and throughout the southeast in all crops. The approximate dollar loss on all crops during 1975 and 1976 in these regions totaled $61.2 and $31.9 million. During 1977 the losses for Georgia alone were approximately $137.5 million for all crops (Sparks 1979).

Fall armyworm will generally feed on foliage, but can also feed on the corn ear during substantial infestations. Feeding will reduce grain weight and overall yield because of foliage and direct ear feeding (Sparks 1979). Small infestations (6%) at growth stages V9 and above did not impact yield of field corn grown in Maryland (Harrison 1984a, 1984b). However, infestations ≥ 63% reduced yield ≥ 20.9%.

Transgenic corn hybrids expressing insecticidal proteins from *Bacillus thuringiensis* were commercially introduced during 1996 (Perlak et al. 2001). Bt corn may express proteins active against coleopteran pests, proteins active against lepidopteran pests, or a combination of proteins to target both groups of pests. Those expressing lepidopteran active proteins were introduced to manage corn borers, including European corn borer, southwestern corn borer, and sugarcane borer, *Diatraea saccharalis* (F.), but have activity against other lepidopteran pests (Estruch et al. 1996; Yu et al. 1997). The lepidopteran active proteins expressed in Bt corn include, Cry1Ab, Cry1F, Cry2Ab, Cry1A.105, and Vip3A. Hybrids may express a single protein; Cry1Ab, ex. YieldGard® corn borer; Cry1F, ex. Herculex® I; or multiple proteins; Cry1Ab+Cry1F, ex. Optimum® Intrasect®; Cry1A.105+Cry2Ab, ex. Genuity® VT Double Pro®;

Cry1F+Cry1A.105+Cry2Ab+Vip3A, ex. Trecepta®. As a component of resistance management plans for Bt corn, a structured refuge of non-Bt corn is required (Catchot et al. 2018). Refuge requirements, such as size and proximity to Bt corn fields, differ somewhat depending on what type of proteins are expressed (coleopteran or lepidopteran active) and geography (cotton growing areas or non-cotton growing areas). The variation in requirements based on geography is due to the same or similar proteins being expressed in both Bt corn and cotton. In cotton growing regions of the United States, a 50% refuge is required for single trait lepidopteran active Bt corn products and a 20% refuge is required for multi lepidopteran active trait products (Monsanto. 2018). Transgenic Bt corn technologies, originally targeting the corn borer complex, have provided good control of fall armyworm. However, resistance to the Cry1F protein has been documented in fall armyworm (Storer et al. 2010, 2012., Xinzhi et al. 2011., Haung et al. 2014). When infestations occur, treatment thresholds are needed to manage fall armyworm infesting non-Bt refuge corn and non-Bt corn planted for general production.

Current fall armyworm thresholds in corn vary by state. In Mississippi, the action threshold is 100% infestation from emergence to the whorl stage (Catchot et al. 2018). In Kentucky, control is warranted when 25% of plants show damage symptoms and live larvae are still present (Bessin 2004). In Georgia, control is recommend when 30% of plants are infested during the whorl stage (Buntin and All 2018). For the state of North Carolina, if severe infestation occurs (50% or greater of plants with fall armyworms) grain yields may be reduced by leaf feeding (Reisig 2018). Tennessee recommends treatment when 50% of plants have one or more larvae per plant (Stewart 2018). In Texas, there is currently no recommended fall armyworm threshold in whorl stage corn (Porter et al. 2011). The overall threshold for fall armyworm in Mid-South whorl stage corn ranges from 25% to 100% plants infested before treatment is warranted, while some states do not have a recommended threshold.

In sorghum, the fall armyworm has been known to feed on the foliage along with the panicles (Henderson et al. 1966). For Mississippi, it is recommended to treat for fall armyworm when there is an average of 75 to 100% of plants infested (Catchot et al. 2018). In Tennessee, one larva per whorl or grain head warrants treatment (Stewart 2018). For Texas, insecticide control is recommended if larval feeding reduces leaf area by more than 30% or is damaging the developing grain head or growing point within the whorl (Cronholm et al. 1998). The threshold for the state of Georgia is 40% infested plants (Buntin and All 2018). In studies conducted by Starks and Burton (1979), they concluded that early infestations of fall armyworm in sorghum did not cause significant yield loss; however, infestations that occur 30 days after planting and later have a higher probability to reduce yield of the sorghum crop. Studies also found that fall armyworm feeding on leaf tissue rarely caused economic losses, but feeding on the sorghum panicles would show yield loss (Teetes and Pendleton. 2000). The overall threshold for fall armyworm in Mid-South whorl stage sorghum ranges from 30% leaf area damaged to 100% of plants infested overall, while some states do not have any recommended threshold.

Treatment thresholds for fall armyworm infesting corn and grain sorghum vary greatly across the Mid-South and Southeastern U.S. With ≥ 20% of the field corn and all of the grain sorghum in Mississippi susceptible to fall armyworm infestations, refined/validated treatment thresholds are needed. These thresholds could become more important if resistance in fall armyworm to Bt trait(s) becomes more prevalent.

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CHAPTER II

IMPACT OF FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE) FEEDING ON YIELD IN WHORL STAGE FIELD CORN

Abstract

Studies were conducted during 2016 and 2017 at Starkville and Stoneville, MS to determine the impact of fall armyworm damage on yield of field corn. Fall armyworm larvae were infested at two vegetative growth stages and five infestation levels. Infestation level had a significant effect on damage, but not on yield. Manual damage conducted at the same growth stages and levels only impacted yield at the V10 growth stage. For every 1% increase in damaged plants, a yield loss of 29.95 kg/ha was observed. An additional study was conducted to evaluate the sensitivity of vegetative growth stages to defoliation. Yield loss was significantly greater when defoliation occurred during the V9 through the V15 growth stages compared to the V3 to V7 growth stages. However, significantly less plant biomass loss was required to impact yield during the V9 to V15 growth stages compared to other growth stages.

Introduction

Field corn, *Zea mays* (L.), is produced mainly for livestock feed, ethanol, cereal products, and human-food products including corn syrup, high fructose corn syrup, corn oil, and corn starch. Corn is the most widely grown field crop in the United States with estimated production of 38,042,069 ha during 2016 and 36,595,317 ha during 2017

(USDA NASS 2016). Production in Mississippi was estimated at 291,373 ha during 2016 and 218,530 ha during 2017 (USDA NASS 2016). Average yields during 2016 and 2017 were 10,900 kg/ha and 12,000 kg/ha respectively with a value of $447,000,000 and $344,000,000, based on a price average of $0.13/kg.

The fall armyworm, *Spodoptera frugiperda* (J.E. Smith), is a polyphagous pest that infests cotton, *Gossypium hirsutum* (L.); corn, *Zea mays* (L.); soybean, *Glycine max* (L.) Merr.; and grain sorghum, *Sorghum bicolor* (L.); among other crops (Hayes 1988). It is a major pest of all plants in the Poaceae family that are found in the United States anywhere east of the Rocky Mountains and as far north as southern Canada (Capinera 1999). The fall armyworm has over sixty host plants, but it prefers to feed on members of the grass family (Poaceae), if they are available. Some of these common grass hosts include: bermudagrass, *Cynodon dactylon* (L.); corn, crabgrass, *Digitaria spp*.; and grain sorghum, (Luginbill 1928). The fall armyworm can only survive winters in the United States in the tropical and subtropical regions such as the southern tips of Florida and Texas. In these areas, temperatures rarely fall below 10° C (Sparks 1979). Some have the ability to cover up to 482 km in a single generation (Ashley et al. 1989). Depending on weather conditions, the fall armyworm will normally reach the northern portion of Mississippi around late-May to June (Sparks 1979).

The fall armyworm has been reported to be found in the whorl of corn plants, and as soon as the plant tassels, it will move to the lower leaves or the ear (Labatte 1993). Fall armyworm feeding is known to cause reduced grain weight and overall yields in part to foliar and direct ear feeding in field corn (Sparks 1979). Larvae are capable of consuming all leaf tissue, except the midrib. In more severe infestations, large numbers of larvae will be reduced to one or two per plant due to cannibalism (Sparks 1979). Symptoms of feeding on corn will include moist sawdust-like frass on the upper leaves and whorl of the plant. When earlier instars are feeding, damage may appear similar to that of European corn borer. However late instar European corn borer larvae will bore into the stalk, while late instar fall armyworm feed on foliage. Corn earworm foliage feeding can also look similar to early instar fall armyworm damage in reference to damage found in the whorl. However when referring to ear feeding, the corn earworm damage includes visible frass and chewed kernels at the ear tip mainly due to eggs being laid in corn silks. Contrarily, the fall armyworm feeds through the husk, but does not always feed down through the silks as the corn earworm does (Bohnenblust et al. 2012). In a study conducted by Mulder and Showers (1986) in Iowa, armyworm, *Pseudaletia unipuncta* (Haworth), larva were infested at various percentage levels, densities, and instars to corn predominantly in 7-8 leaf stage and 9-10 leaf stage. They reported no differences in yield across the densities infested.

Transgenic corn hybrids expressing insecticidal proteins from *Bacillus thuringiensis* were commercially introduced during 1996 (Perlak et al. 2001). Bt corn may express proteins active against coleopteran pests, proteins active against lepidopteran pests, or a combination of proteins to target both groups of pests. Adoption of Bt corn for coleopteran and lepidopteran insect pest management has increased from 8% in 1997 to 81% in 2015 (USDA 2017). Those expressing lepidopteran active proteins were introduced to manage corn borers, including European corn borer, *Ostrinia nubilalis* (Hübner), southwestern corn borer, *Diatraea* grandiosella (Dyar), and sugarcane borer, *Diatraea saccharalis* (F.), but have activity against other lepidopteran pests (Estruch et al. 1996; Yu et al. 1997). The lepidopteran active proteins expressed in Bt corn include, Cry1Ab, Cry1F, Cry2Ab, Cry1A.105, and Vip3A. Hybrids may express a single protein; Cry1Ab, ex. YieldGard® corn borer; Cry1F, ex. Herculex® I; or multiple proteins; Cry1Ab+Cry1F, ex. Optimum® Intrasect®; Cry1A.105+Cry2Ab, ex. Genuity® VT Double Pro®; Cry1F+Cry1A.105+Cry2Ab+Vip3A, ex. Trecepta®. As a component of resistance management plans for Bt corn, a structured refuge of non-Bt corn is required (Catchot et al. 2018). Refuge requirements, such as size and proximity to Bt corn fields, differ somewhat depending on what type of proteins are expressed (coleopteran or lepidopteran active) and geography (cotton growing areas or non-cotton growing areas). The variation in requirements based on geography is due to the same or similar proteins being expressed in both Bt corn and cotton. In cotton growing regions of the United States, a 50% refuge is required for single trait lepidopteran active Bt corn products and a 20% refuge is required for multi lepidopteran active trait products (Catchot et al. 2018). Transgenic Bt corn technologies, originally targeting the corn borer complex, have provided good control of fall armyworm. However, resistance to the Cry1F protein has been documented in fall armyworm (Storer et al. 2010, 2012., Xinzhi et al. 2011., Haung et al. 2014).

Thresholds for whorl stage corn vary significantly throughout the Mid-South region. Some states have no recommended threshold, while others recommend treatment when 25% to 100% of plants are infested. With refuge corn and non-Bt corn for general production, ≥ 20% of the field corn in Mississippi is susceptible to fall armyworm infestations. Refined/validated treatment thresholds are needed and could become more important if resistance to Bt trait(s) becomes more prevalent.

Materials and Methods

To determine the impact of defoliation and fall armyworm infestations during vegetative growth stages on field corn yield, experiments were conducted at the Mississippi State University Delta Research & Extension Center in Stoneville, MS and at the Mississippi State University R. R. Foil Research Farm in Starkville, MS during 2016 and 2017. These experiments were conducted at the V5 and V10 growth stages (Abendroth et al. 2011). These growth stages were chosen based on observations of natural fall armyworm infestations in Mississippi. Typically initial infestations of fall armyworm in later planted corn have been observed during the V4-V6 growth stages. While initial infestations in earlier planted corn typically occur during the V9 to V11 growth stages. Two separate experiments were conducted at the previously mentioned growth stages. One study used artificial infestations of fall armyworm larvae, while the other experiment utilized manual damage methods. For both experiments, the experimental design was a randomized complete block with four replications. Plot size for experiments at the Starkville location was four rows (96.5 cm centers) by 3.05 m in length. Plots were planted at a seeding rate of 77,500 seed/ha. At the Stoneville location, plot size was four rows (101.6 cm centers) by 3.05 m in length, with a seeding rate of 85,000 seed/ha.

For the infestation studies, Dekalb DKC67-70RR2 (non-Bt) corn seed (Monsanto Company, St. Louis, MO) was utilized. Planting dates for these studies ranged from mid Mar to mid Apr during each year. Each study (V5 and V10) were repeated eight times across locations and years. The treatments in these studies consisted of 0%, 25%, 50%, 75%, or 100% of plants on the center two rows of each plot infested. All of the plants on the center two rows of each plot were counted to determine the appropriate number of plants to infest. Plants to be infested were selected at random and marked with a wire pin flag for later identification. These plants were infested with 15-20 neonate fall armyworm larvae using a Davis inoculator and methods described in Davis and Oswalt (1979). Plots were monitored and when larvae had left the plant to pupate, feeding damage was estimated using the rating scale described by Davis et al. (1992), which is outlined in Table 2.1. Plants were rated individually and mean damage per plot and mean damage per infested plant was calculated. When grain reached a harvestable moisture content, plots were mechanically harvested. Grain yields were converted to kg/ha at a standard moisture content of 15%.

During 2016, larvae from a laboratory colony of fall armyworm were used to infest plots. During 2017, larvae from a fall armyworm colony collected from non-Bt corn during Mar 2017 were utilized. Subsets of the fall armyworm colonies were maintained at the insect rearing facilities at Mississippi State University (Starkville) and at the Mississippi State University Delta Research & Extension Center (Stoneville) under conditions of 25°C, 80% relative humidity, and 16:8 (L:D) photoperiod. Adults were maintained in 3.79 liter cardboard containers (ca. 50-75 per container) lined with waxed paper and covered with cheese cloth, both served as oviposition substrates. Adults were fed a 10% beer/honey water solution. Egg sheets (cheese cloth and wax paper) were collected daily and were placed in 3.79 liter Ziploc® (S.C Johnson & Johnson, Inc., Racine WI) bags until larval eclosion. Larvae were either used for infestations or to continue the colony. Larvae retained for colony maintenance were placed in 29.5 ml plastic cups (Solo Co., Urbana, IL) containing Stonefly Heliothis Diet (Product No. 38-0600, Ward's Natural Science, Rochester, NY) with matching lids.

For the manual damage studies, Dekalb DKC67-72VT2P corn seed (Monsanto Company, St. Louis, MO) was utilized. This hybrid expresses the Cry1A.105 and Cry2Ab Bt proteins and was utilized to minimize damage from natural infestations of lepidopteran insect pests. Planting dates for these studies ranged from mid Mar to mid Apr during each year. Each study (V5 and V10) were repeated seven times across locations and years. The treatments in these studies consisted of removal of all plant tissue at and above the upper most fully expanded leaf collar from 0, 25, 50, 75, or 100% of plants on the center two rows of each plot. All of the plants on the center two rows of each plot were counted to determine the appropriate number of plants to be damaged. Plant material was removed with garden shears. To estimate the amount of above ground tissue removed, 25 plants were randomly selected from outside rows of plots across the trial area. Plants were cut at the soil surface and transported to the laboratory. Plants were partitioned into the portion at and above the upper most expanded leaf collar and the portion below the upper most expanded leaf collar. Weights of each plant portion were determined. Mean percent of above ground biomass removed in the studies conducted at V5 was 40.8%, while mean percent of above ground biomass removed in the studies conducted at V10 was 33.8%. Plant heights were taken at each growth stage. Mean height of the above ground portion of the plant conducted at V5 was 30.22 cm, while mean height of the above ground portion of the plant conducted at V10 was 53.08 cm. When grain reached a harvestable moisture content, plots were mechanically harvested. Grain yields were converted to kg/ha at a standard moisture content of 15%.

For the infestation studies, mean damage rating per plot was subjected to analysis of variance (PROC GLIMMIX, SAS Institute 2011). Infestation level was considered a fixed effect and replication, location, year, and replication nested in location by year as random effects. Degrees of freedom were calculated using the Kenward-Roger method. Means were separated according to Fisher's Protected Least Significant Difference with α of 0.05. For both the infestation and manual damage studies, yield data were subjected to regression analysis (PROC GLIMMIX, SAS Institute 2011). In the infestation studies, natural infestation and/or interplant movement of larvae occurred, therefore the percentage of plants with fall armyworm damage was used in the regression analysis instead of the percentage of plants infested. Infestation or damage level was considered a fixed effect and replication, location, year, and replication nested in location by year as random effects. Degrees of freedom were calculated using the Kenward-Roger method.

To determine the sensitivity of field corn across a range of vegetative growth stages to defoliation, an experiment was conducted at the Mississippi State University Delta Research and Extension Center in Stoneville, MS and the Mississippi State University R.R. Foil Research Farm in Starkville, MS during 2017 using manual damage methods. Plot size and seeding rates for each location were the same as described for the previous experiments. Planting dates for these studies ranged from mid Mar to mid Apr. Dekalb DKC67-72VT2P corn seed (Monsanto Company, St. Louis, MO) was utilized. This hybrid expresses the Cry1A.105 and Cry2Ab Bt proteins and was utilized to minimize damage from natural infestations of lepidopteran insect pests. Treatments were arranged in a randomized complete block design with four replications and included removal of all plant tissue at and above the upper most fully expanded leaf collar from all plants on the center two rows of each plot at the V3, V5, V7, V9, V11, V13, V15, or V17, and an undamaged control. Plant tissue removal procedures were the same as described above. This study was repeated five times across both locations. The percentage of above ground biomass removed at each growth stage was determined using the methods described in the previous experiment. Percent biomass removed data were subjected to analysis of variance (PROC GLIMMIX, SAS Institute 2011). Growth stage was considered a fixed effect and replication as a random effect. Because the percent biomass removed at each growth stage was determined by collecting one sample of 25 plants per growth stage per trial. Individual trials were considered as replicates in the analysis. Degrees of freedom were calculated using the Kenward-Roger method. Means were separated according to Fisher's Protected Least Significant Difference with α of 0.05. When grain reached a harvestable moisture content, plots were mechanically harvested. Grain yields were converted to kg/ha at a standard moisture content of 15%. Data were subjected to analysis of variance (PROC GLIMMIX, SAS Institute 2011). Growth stage was considered a fixed effect and replication, location, and replication nested in location as random effects. Degrees of freedom were calculated using the Kenward-Roger method. Means were separated according to Fisher's Protected Least Significant Difference with α of 0.05.

Results

For infestation studies conducted at the V5 growth stage, some natural infestations and/or interplant movement of larvae occurred (Fig. 2.1). The 25%, 50%, 75% infested plots and the non-infested plots had higher percentages of plants with fall armyworm damage than the percentage of plants that were infested. The 100% infested plots (85.6% damaged plants) had significantly higher percent damaged plants than all other plots, except the plots with 75% plants manually infested ($F=81.99$; df 4,110; $P<0.01$) (Fig. 2.1). Also, the 50% and 75% infested plots had significantly higher percent damaged plants than the 25% infested plots and the non-infested plots. The non-infested plots had a significantly lower percentage of plants (10.3%) with fall armyworm damage than all other plots. There were significant differences among infestation levels for mean damage rating per plot ($F=48.68$; df 4,110; $P<0.01$) (Fig. 2.2). The 100% infested plots had a significantly higher mean damage rating compared to the 25% and 50% infested plots, with the 50% infested plots also having a significantly higher per plot damage ratings than the 25% infested plots. The non-infested plots had significantly lower mean damage rating per plot compared all of the infested plots. There was no significant relationship between percentage of plants with fall armyworm damage and yield ($F=0.99$; df 1,99; $P=0.32$) (Fig. 2.3).

For infestation studies conducted at the V10 growth stage, natural infestations and/or interplant movement of larvae occurred. There were significant differences in the percentage of damaged plants among infestation levels ($F=157.21$; df 4,156.1; $P<0.01$) (Fig. 2.4). The 100% infested plots (93.3% damaged plants) had significantly higher percent damaged plants than all other plots. Also, the 50% and 75% infested plots had significantly higher percent damaged plants than the 25% infested plots and the non-infested plots. The non-infested plots had a significantly lower percentage of plants (9.7%) with fall armyworm damage than all other plots. There were significant differences for mean damage rating per plot ($F=112.31$; df 4, 156; $P<0.01$) (Fig. 2.5). The 100% infested plots had a significantly higher mean damage rating compared to plots infested at any of the other levels. Also, the 50% and 75% infested plots had significantly higher per plot damage ratings than the 25% infested plots. The non-infested plots had significantly lower mean damage rating per plot compared to all of the infested plots. There was no significant relationship between percentage of plants with fall armyworm damage and yield ($F=2.19$; df 1, 135; $P=0.14$) (Fig. 2.6).

Manual damage studies were also conducted at the V5 and V10 growth stages. With removal of 40.8% of the above ground biomass/plant at the V5 growth stage, there was no significant relationship between percent damaged plants and yield ($F=1.82$; df 1,111; $P=0.18$) (Fig. 2.7). When damage was imposed at the V10 growth stage, a significant relationship between percent damaged plants and yield was observed ($F=32.66$; df 1,119; $P<0.01$) with a mean removal of 33.8% of the above ground biomass. For every 1% increase in damaged plants a yield loss of 29.95 kg/ha was observed (Fig 2.8).

An additional study was conducted to evaluate the sensitivity of vegetative growth stages of field corn to defoliation. There was a significant difference in the amount of above ground biomass removed at the various growth stages of field corn ($F=15.43$; df 7, 32; $P<0.01$) (Fig. 2.9). Significantly more above ground biomass was removed at the V3 and V5 growth stages compared to growth stage of V9 to V17. Yield was significantly reduced when defoliation occurred during the V9 to V15 growth stages compared to the V3 to V7 growth stages, the V17 growth stage, and the non-damaged control ($F=9.1$; df 8,152; $P<0.01$) (Fig. 2.10). Yield loss was significantly greater with defoliation during the V9 through the V15 growth stages compared to the V3 to V7 growth stages even though significantly less biomass was removed. Additionally, significantly less defoliation was required to reduce yield at the V13 and V15 growth stages compared to the V9 growth stages.

Discussion

Manual removal of ca. 36%-41% of the above ground biomass from 100% of plants did not impact yield during the V3-V7 growth stages. Also, 100% of plants infested with fall armyworm larvae at the V5 growth stage did not impact yield. Based on these studies it does not appear that fall armyworm infestations during the V3-V7 growth stages are economically important for corn planted during the recommended planting window in Mississippi. These results are similar to those of Mulder and Showers (1982) in which feeding by the true armyworm (*Psuedaletia unipuncta*) (Haworth) during the V7-V10 may have little influence on corn growth or yields (Mulder and Showers 1986). However the impact of infestations that result in higher levels of defoliation that could occur with later plantings may be important. Further research is needed to determine the impact of greater levels of defoliation occurring during these early vegetative growth stages. Manual removal of ca. 34% of above ground biomass at the V10 growth stage impacted yield. However, fall armyworm infestations at the same growth stage did not cause enough damage to impact yield. Additional manual damage studies indicated that the V9 to V15 growth stages are sensitive to defoliation at the levels of removal included in the study. Also, the current study indicates that the sensitivity of field corn to defoliation increases from the V9 to V15 growth stages as significantly less defoliation was required to reduce yield at V13 and V15 compared to the V9 growth stage. These results concur with those of Klein and Shapiro (2011) during their study on hail damage to various growth stages of corn. Their studies demonstrated that as the growth stage at which damaged occurred increased, the amount of defoliation needed to reduce yield decreases. This indicates that treatment may need to be considered when infestations occur during these growth stages. There is a lack of lepidopteran defoliation studies during later vegetative growth stages corn. Because of this, additional research is needed to determine the critical levels of infestation and defoliation to impact yield at specific growth stages.

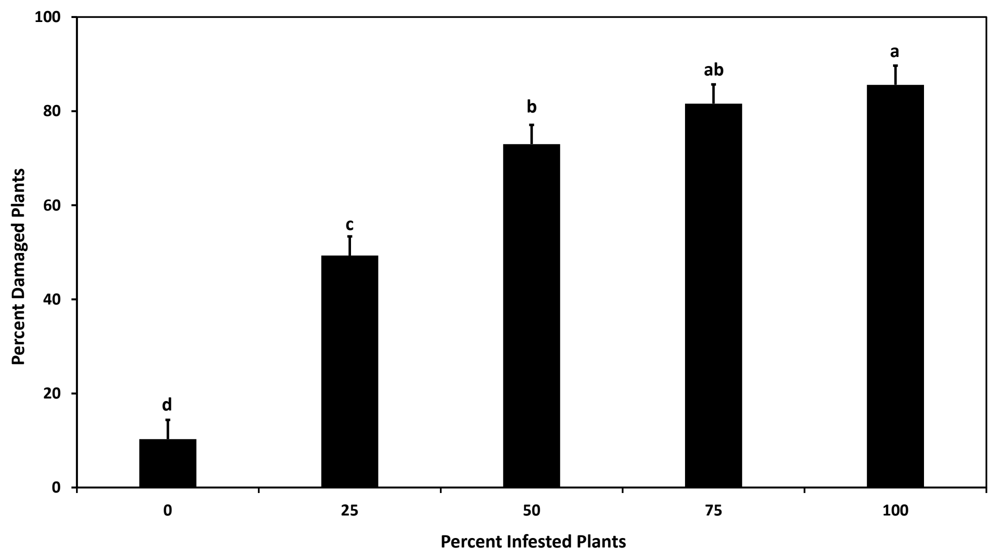

Figure 2.1 Mean (SE) percentage of field corn, *Zea mays*, with fall armyworm, *Spodoptera frugiperda*, feeding damage from infestations conducted at the V5 growth stage in Stoneville and Starkville, MS in 2016 and 2017. Means with a common letter are not different (Fisher's PLSD, P≤0.05).

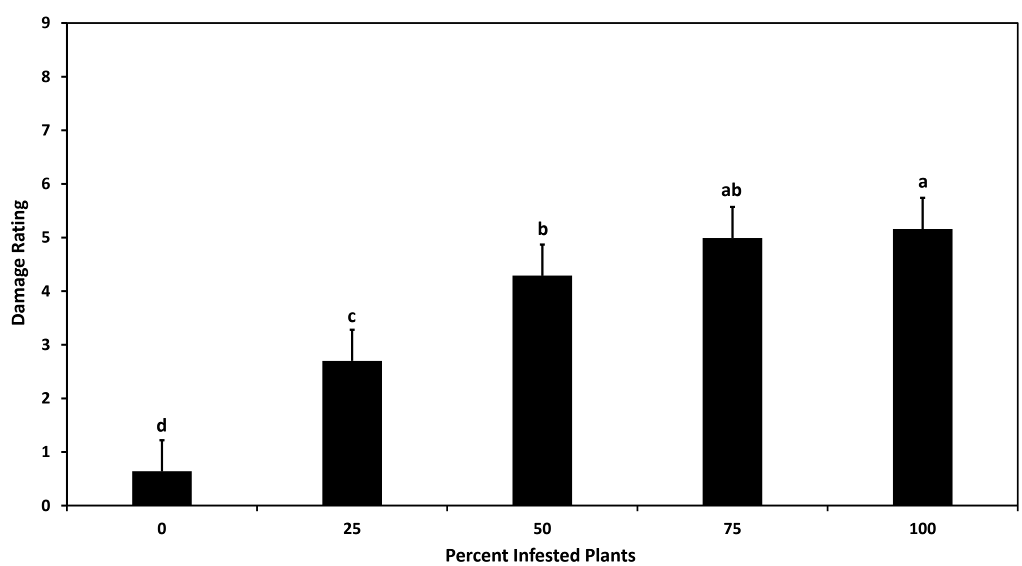

Figure 2.2 Mean (SE) damage rating per plot for artificial infestations to field corn, *Zea mays*, with fall armyworm, *Spodoptera frugiperda*, conducted at the V5 growth stage in Stoneville and Starkville, MS in 2016 and 2017. Bars with a common letter are not significantly different. (Fisher's PLSD, P≤0.05).

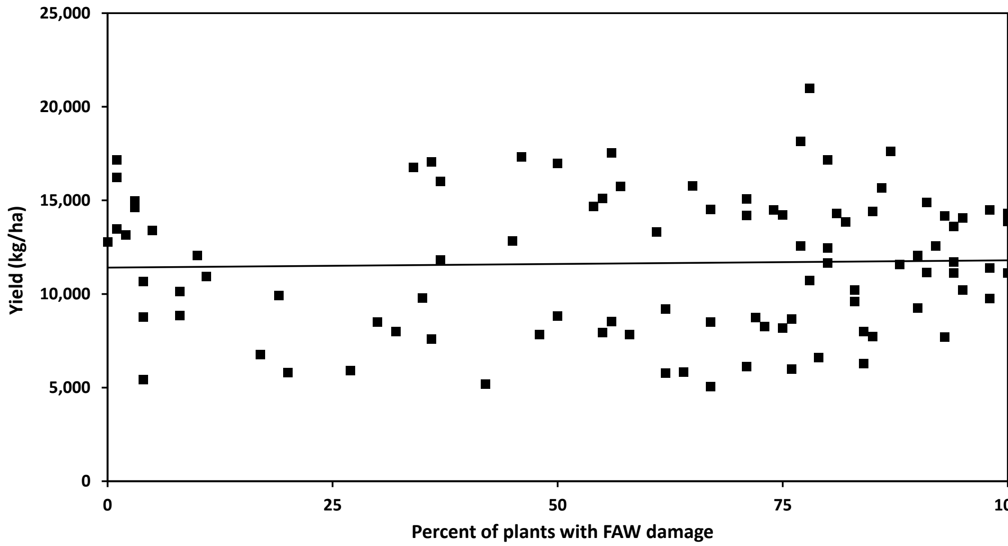

Figure 2.3 Relationship between percent of plants with fall armyworm, *Spodoptera frugiperda*, damage and field corn, *Zea mays*, yield (P=0.3214; kg/ha=12,041-5.008x (±5.02)). Artificial infestations conducted at the V5 growth stage in Stoneville and Starkville, MS in 2016 and 2017.

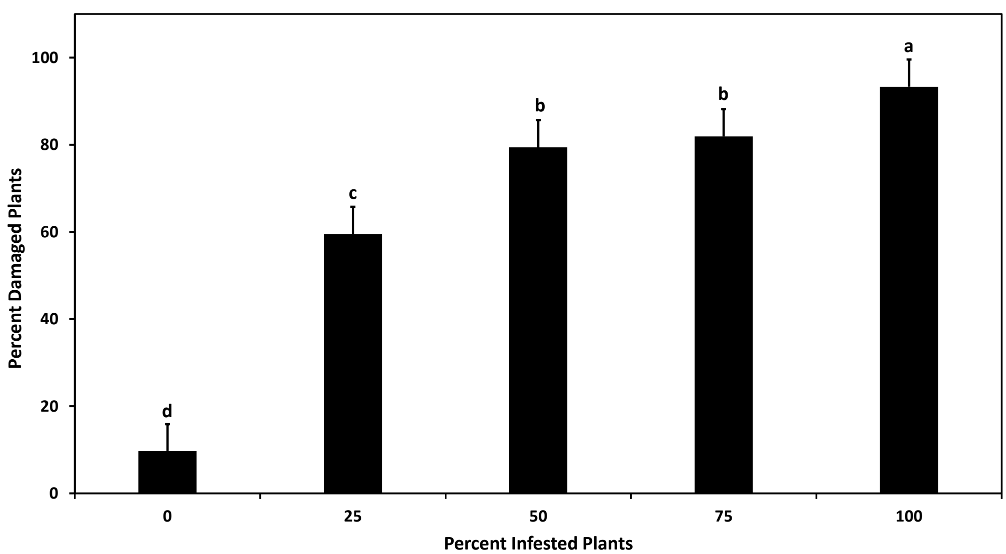

Figure 2.4 Mean (SE) percentage of field corn, *Zea mays*, with fall armyworm, *Spodoptera frugiperda*, feeding damage from infestations conducted at the V10 growth stage in Stoneville and Starkville, MS in 2016 and 2017. Means with a common letter are not significantly different (Fisher's PLSD, $P \leq 0.05$).

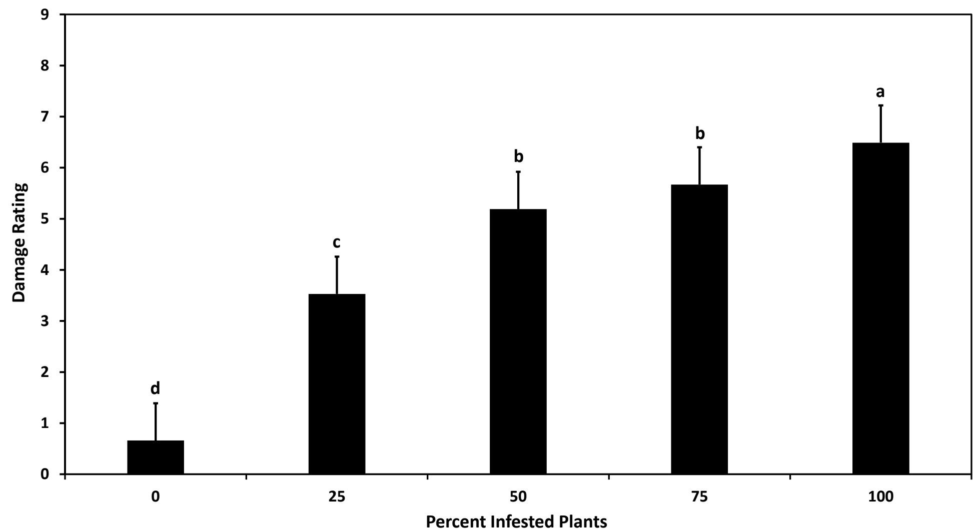

Figure 2.5 Mean (SE) damage rating per plot for artificial infestations to field corn, *Zea mays*, with fall armyworm, *Spodoptera frugiperda*, conducted at the V10 growth stage in Stoneville and Starkville, MS in 2016 and 2017. Bars with a common letter are not significantly different. (Fisher's PLSD, $P \leq 0.05$).

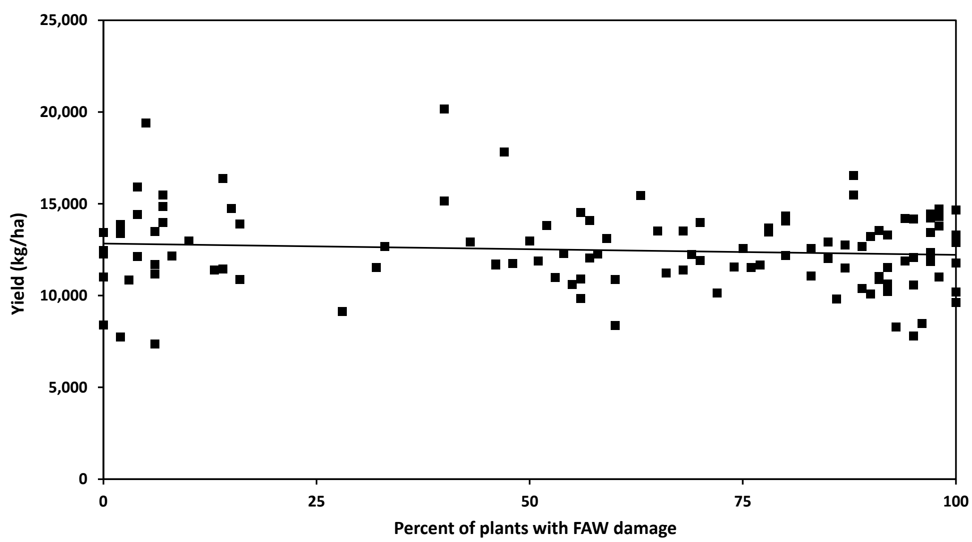

Figure 2.6 Relationship between percent of plants with fall armyworm, *Spodoptera frugiperda*, damage and field corn, *Zea mays*, yield (P=0.1408; kg/ha=12,921-6.56x (±4.43)). Artificial infestations conducted at the V10 growth stage in Stoneville and Starkville, MS in 2016 and 2017.

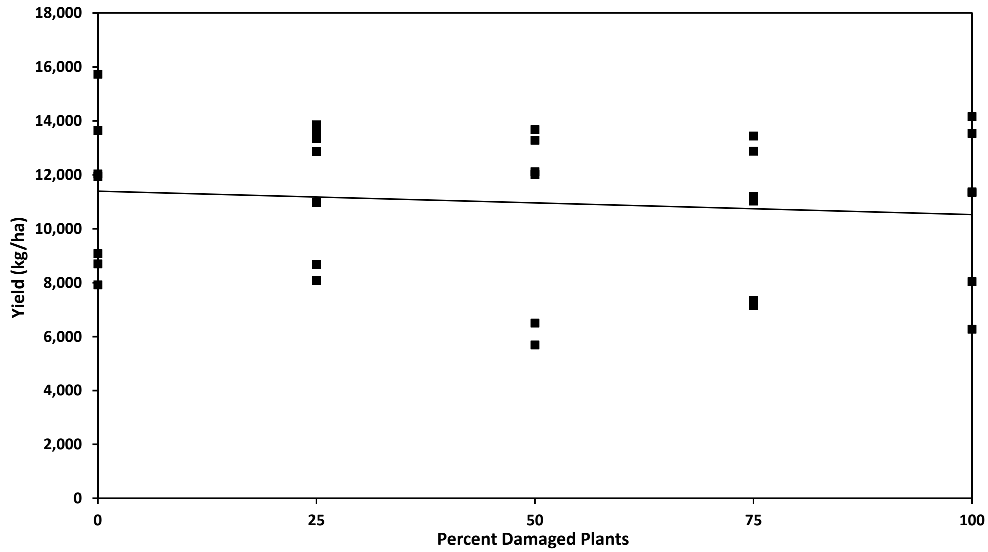

Figure 2.7 Relationship between percent damaged plants and field corn, *Zea mays*, yield (P=0.18; kg/ha=11,352-7.41x (±5.49)). Manual damage treatments imposed at the V5 growth stage in Stoneville and Starkville, MS in 2016 and 2017.

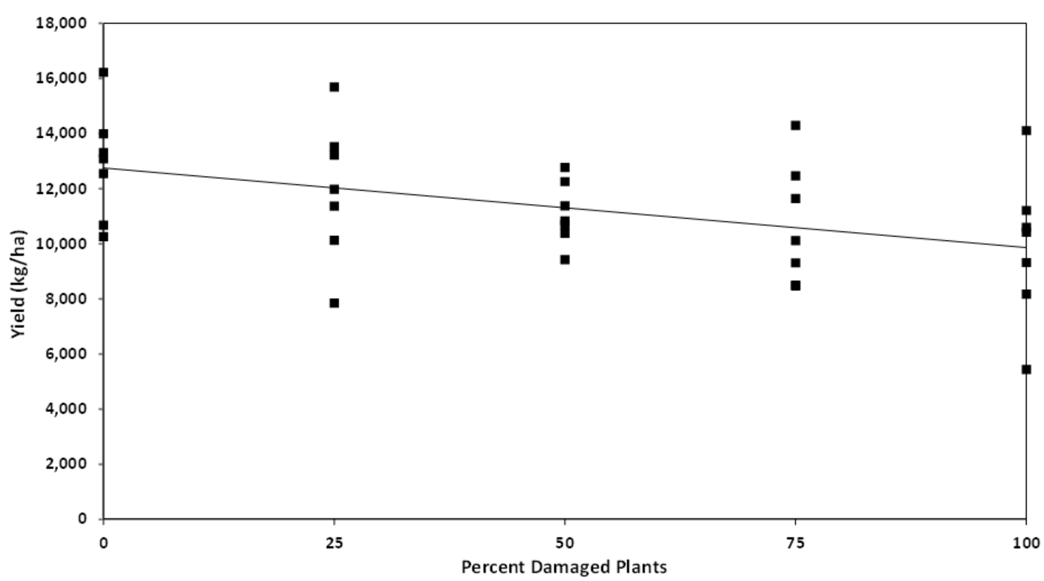

Figure 2.8 Relationship between percent damaged plants and field corn, *Zea mays*, yield (P<0.01; kg/ha=12,823-29.95x (±5.24)). Manual damage treatments imposed at the V10 growth stage in Stoneville and Starkville, MS in 2016 and 2017.

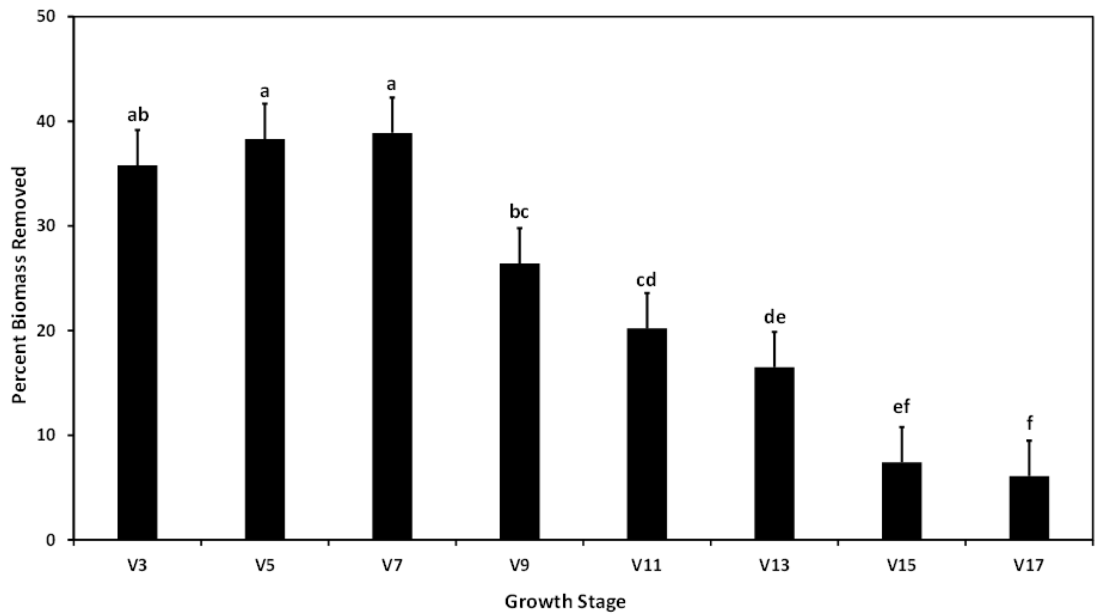

Figure 2.9 Percent of above ground biomass removed from field corn, *Zea mays*, in Stoneville and Starkville, MS in 2017. Bars with a common letter are not significantly different (Fisher's PLSD, P≤0.05).

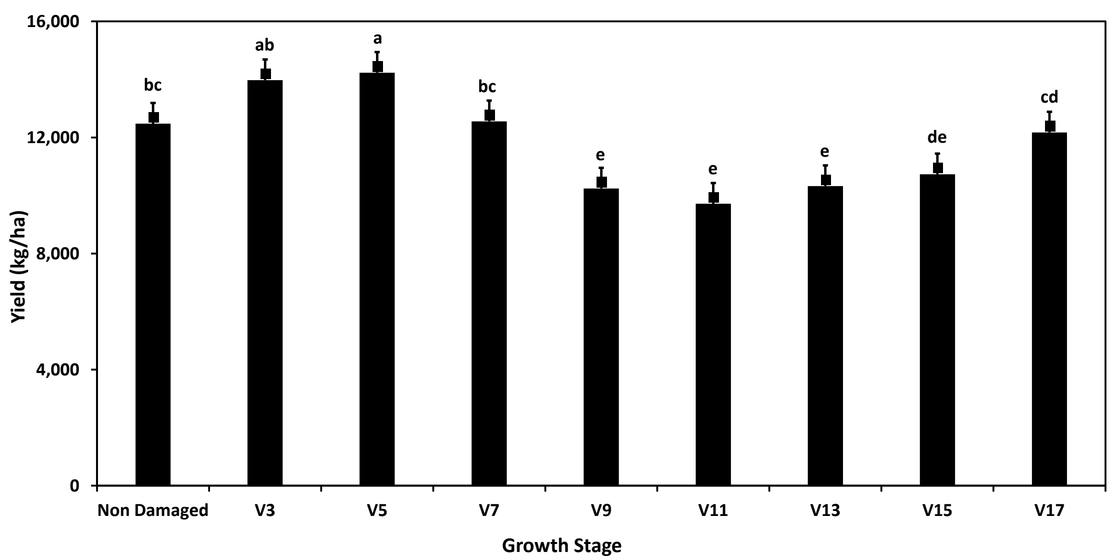

Figure 2.10 Impact of manual defoliation at selected vegetative growth stages on field corn, *Zea mays*, yield in Stoneville and Starkville, MS in 2017. Bars with a common letter are not significantly different (P≤0.05, Fisher's PLSD).

Table 2.1 Damage scale used for damage assessment as described by Davis et al. (1992).

Score	Description
0	**No visible damage.**
1	**Only pinhole lesions present on whorl leaves.**
2	**Pinholes and small circular lesions present on whorl leaves.**
3	**Pinholes, small circular lesions and few elongated lesions of up to 1.3 cm in length present on whorl and/or furl leaves.**
4	**Small elongated lesions present on whorl leaves and a few mid-size elongated lesions of 1.3 to 2.5 cm in length present on whorl and/or furl leaves.**
5	**Small elongated lesions and several mid-size elongated lesions present on whorl and furl leaves.**
6	**Small and mid-size elongated lesions plus a few large elongated lesions of greater than 2.5 cm in length present on whorl and/or furl leaves.**
7	**Many small and mid-size elongated lesions present on whorl leaves plus several large elongated lesions present on the furl leaves.**
8	**Many small and mid-size elongated lesions present on whorl leaves plus many large elongated lesions present on the furl leaves.**
9	**Many elongated lesions of all sizes on whorl and furl leaves plus a few uniform to irregular shaped holes (basement membrane consumed) eaten from the base of the whorl and/or furl leaves.**

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CHAPTER III

IMPACT OF FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE) FEEDING ON YIELD IN WHORL STAGE GRAIN SORGHUM

Abstract

Research studies were conducted during 2017 in Starkville and Stoneville, MS to determine the impact of fall armyworm, *Spodoptera frugiperda* (J.E. Smith), on damage and yield of grain sorghum, *Sorghum bicolor* (L.) Moench. Fall armyworm larvae were infested at the V7 vegetative growth stage and five infestation levels. Infestation level had a significant effect on damage, but not on yield. Manual damage conducted at the same growth stage and infestation/damage level did not have a significant effect on yield at the V7 growth stage. An additional study was conducted to evaluate the sensitivity of vegetative growth stages to defoliation. Yield was significantly reduced when defoliation occurred during the V9 growth stage compared to the V5 growth stage. However, significantly more above ground biomass was removed at the V3 and V5 growth stages compared to growth stages V7 and V9.

Introduction

Grain sorghum, *Sorghum bicolor* (L.) Moench, is one of the top five cereal grain crops that is produced worldwide. Of all of the countries that produce grain sorghum, the United States is the leader in hectares grown and kilograms produced. Approximately 2.5 million hectares of grain sorghum were harvested in the United States during 2016, with a slight decrease in 2017 to only 2.3 million hectares (USDA NASS 2016). Production in Mississippi was estimated at 4,400 ha during 2016 and 3,600 ha during 2017 (USDA NASS 2016). Average yields during 2016 and 2017 were 5,600 kg/ha with a value of $3,218,000 and $968,000, respectively, based on a price average of $0.12/kg and $0.13/kg, respectively. A benefit of producing grain sorghum is its ability to tolerate heat and drought relatively well. It requires much less water to produce a grain yield at full potential (Rosenow et al. 1983). Grain sorghum can also be used as a rotation crop in order to help with weed and pest management (Cronholm et al. 1998).

The fall armyworm, *Spodoptera frugiperda* (J.E. Smith), is a major pest of the Poaceae family and it is found throughout the eastern United States including areas as far west as the Rocky Mountains and as far north as the southern part of Canada (Capinera 1999, Sparks 1979, Ashley et al. 1989). Even though the fall armyworm has over 60 host plants recognized, it prefers to feed on members of the (Poaceae) grass family, if they are available. Some of the most preferred grass hosts include: bermudagrass, *Cynodon dactylon* (L.); corn, *Zea mays* (L.); crabgrass, *Digitaria spp*; and grain sorghum, *Sorghum bicolor* (L.), (Luginbill 1928).

The fall armyworm is more commonly a pest of later planted grain sorghum in Mississippi (Henderson et al. 1966, Wiseman et al. 1986). Larvae can cause damage to grain sorghum during three important growth stages. The first of these stages is newly emerged seedlings, however, because the growing point is still below the ground, economic damage will seldom occur. The latest stage where fall armyworm can cause damage to grain sorghum is during the grain development stage. During this stage, fall armyworm larvae will consume developing kernels of individual sorghum heads.

This will result in a direct yield loss (Martin et al. 1980). The intermediate stage where damage can occur is the whorl-stage. During the whorl-stage, fall armyworm larvae feed on foliage and therefore reduce the leaf surface area that is absorbing light for photosynthesis. At this stage, the fall armyworm can also destroy the growing point and inhibit further growth of the overall plant. If heavy infestations are encountered during the whorl stage, an increase in tillering, decrease in plant height, and a reduction in grain yield can occur (Henderson et al. 1966, McMillian and Stark 1967, Starks and Burton 1979).

Fall armyworm larvae will cause substantially more damage as they get larger, with ca. 80% of the direct damage to grain caused by fifth and sixth-instar larvae (Teetes and Pendleton 2000). While fall armyworm is known to be a cannibalistic insect, it is not as cannibalistic as others. Because of this, more sorghum larvae per individual sorghum panicle can be found (Teetes and Pendleton 2000). This can allow large populations of large larvae to cause significant damage and result in severe yield loss (Teetes and Pendleton 2000).

Thresholds for whorl stage sorghum vary significantly throughout the Mid-South region. Some states have no recommended threshold, while others recommend treatment when 30% of leaf tissue is removed on each plant to 100% of plants being infested overall. With substantial variation in thresholds across the Southern U.S., refined/validated thresholds are needed.

Materials and Methods

To determine the impact of defoliation and fall armyworm infestations during vegetative growth stages on sorghum yield, experiments were conducted at the

Mississippi State University Delta Research & Extension Center in Stoneville, MS and at the Mississippi State University R. R. Foil Research Farm in Starkville, MS during 2017. These experiments were conducted at the V7 growth stage. This growth stage was chosen based on observations of natural fall armyworm infestations in Mississippi. Two separate experiments were conducted at the previously mentioned growth stage. One study used artificial infestations of fall armyworm larvae, while the other experiment utilized manual damage methods. For both experiments the experimental design was a randomized complete block with four replications. Plot size for experiments at the Starkville location was four rows (96.5 cm centers) by 1.52 m in length. Plots were planted at a seeding rate of 198,000 seed/ha. At the Stoneville location, plot size was four rows (101.6 cm centers) by 1.52 m in length, with a seeding rate of 198,000 seed/ha.

For the infestation studies, Hybrid Pioneer™ 84P80 sorghum seed were utilized. Planting dates for these studies ranged from mid-May to mid-Jun during each year. Each study (V7) was repeated four times across locations. The treatments in these studies consisted of 0, 25, 50, 75, or 100% of plants on the center two rows of each plot infested. All of the plants on the center two rows of each plot were counted to determine the appropriate number of plants to infest. Plants to be infested were selected at random and marked with a wire pin flag for later identification. These plants were infested with 15-20 neonate fall armyworm larvae using a Davis inoculator and methods described in Davis and Oswalt (1979). Plots were monitored and when larvae had left the plant to pupate, feeding damage was estimated using the rating scale described by Davis et al. (1992), which is outlined in Table 3.1. Plants were rated individually and mean damage per plot and mean damage per infested plant was calculated. When grain reached a harvestable moisture content, plots were mechanically harvested. Grain yields were converted to kg/ha at a standard moisture content of 15%.

During 2017, larvae from a fall armyworm colony collected from non-Bt corn during Mar 2017 were utilized. Subsets of the fall armyworm colonies were maintained at the insect rearing facilities at Mississippi State University (Starkville) and at the MSU Delta Research & Extension Center (Stoneville) under conditions of 25°C, 80% relative humidity, and 16:8 (L:D) photoperiod. Adults were maintain in 3.79 liter cardboard containers (ca. 50-75 per container) lined with waxed paper and covered with cheese cloth, both served as oviposition substrates. Adults were fed a 10% beer/honey water solution. Egg sheets (cheese cloth and wax paper) were collected daily and were placed in 3.79 liter Ziploc® (S.C Johnson & Johnson, Inc., Racine WI) bags until larval eclosion. Larvae were either used for infestations or to continue the colony. Larvae retained for colony maintenance placed in 29.5 ml plastic cups (Solo Co., Urbana, IL) containing Stonefly Heliothis Diet (Product No. 38-0600, Ward's Natural Science, Rochester, NY) with matching lids.

For the manual damage studies, Hybrid Pioneer™ 84P80 sorghum seed was utilized. Planting dates for these studies ranged from mid-May to mid-Jun during each year. Each study (V7) was repeated four times across locations. The treatments in these studies consisted of removal of all plant tissue at and above the upper most fully expanded leaf collar from 0, 25, 50, 75, or 100% of plants on the center two rows of each plot. All of the plants on the center two rows of each plot were counted to determine the appropriate number of plants to be damaged. Plant material was removed with garden shears. To estimate the amount of above ground tissue removed, 25 plants were randomly selected from outside rows of plots across the trial area. Plants were cut at the soil surface and transported to the laboratory. Plants were partitioned into the portion at and above the upper most expanded leaf collar and the portion below the upper most expanded leaf collar. Weights of each plant portion were determined. Mean percent of above ground biomass removed in the studies conducted at the V7 growth stage was 23.8%. When grain reached a harvestable moisture content, plots were mechanically harvested. Grain yields were converted to kg/ha at a standard moisture content of 15%.

For the infestation studies, mean damage rating per plot was subjected to analysis of variance (PROC GLIMMIX, SAS Institute 2011). Infestation level was considered a fixed effect and replication, location, and replication nested in location as random effects. Degrees of freedom were calculated using the Kenward-Roger method. Means were separated according to Fisher's Protected Least Significant Difference with α of 0.05. For both the infestation and manual damage studies, yield data were subjected to regression analysis (PROC GLIMMIX, SAS Institute 2011). In the infestation studies, natural infestation and/or interplant movement of larvae occurred, therefore the percentage of plants with fall armyworm damage was used in the regression analysis instead of the percentage of plants infested. Infestation or damage level was considered a fixed effect and replication, location, and replication nested in location as random effects. Degrees of freedom were calculated using the Kenward-Roger method.

To determine the sensitivity of sorghum across a range of vegetative growth stages to defoliation, an experiment was conducted at the Mississippi State University Delta Research and Extension Center in Stoneville, MS and the Mississippi State University R.R. Foil Research Farm in Starkville, MS during 2017 using manual damage methods. Plot size and seeding rates for each location were the same as described for the previous experiments. Planting dates for these studies ranged from mid-May to mid-Jun. Hybrid Pioneer™ 84P80 sorghum seed were utilized. Treatments were arranged in a randomized complete block design with four replications and included removal of all plant tissue at and above the upper most fully expanded leaf collar from all plants on the center two rows of each plot at the V3, V5, V7, V9 growth stages, and an undamaged control. Plant tissue removal procedures were the same as described above. This study was repeated four times across both locations. The percentage of above ground biomass removed at each growth stage was determined using the methods described in the previous experiment. Percent biomass removed data were subjected to analysis of variance (PROC GLIMMIX, SAS Institute 2011). Growth stage was considered a fixed effect and replication as a random effect. Because the percent biomass removed at each growth stage was determined by collecting one sample of 25 plants per growth stage per trial. Individual trials were considered as replicates in the analysis. Degrees of freedom were calculated using the Kenward-Roger method. Means were separated according to Fisher's Protected Least Significant Difference with α of 0.05. When grain reached a harvestable moisture content, plots were mechanically harvested. Grain yields were converted to kg/ha at a standard moisture content of 15%. Data were subjected to analysis of variance (PROC GLIMMIX, SAS Institute 2011). Growth stage was considered a fixed effect and replication, location, and replication nested in location as random effects. Degrees of freedom were calculated using the Kenward-Roger method. Means were separated according to Fisher's Protected Least Significant Difference with α of 0.05.

Results

For infestation studies conducted at the V7 growth stage, some natural infestations and/or interplant movement of larvae occurred (Fig 3.1). The non-infested plots and the 25% infested plots had a percentage of plants with fall armyworm damage that was higher than the percentage of plants that were infested. The 100% infested plots (59.6% damaged plants) had significantly higher percent damaged plants than all other plots ($F=5.12$; df 4, 60; $P<0.01$) (Fig 3.1). Also, there were no significant differences between the 25%, 50%, and 75% infested plots and the non-infested plots. There were significant differences among infestation levels for mean damage rating per plot ($F=3.23$; df 4, 75; $P=0.02$). The 100% infested plots had a significantly higher mean damage rating compared to the 25%, 50%, or 75% infested plots or the non-infested plots (Fig 3.2). There was no significant relationship between percentage of plants with fall armyworm damage and yield ($F=1.49$; df 1, 63; $P=0.23$) (Fig 3.3).

Manual damage studies were also conducted at the V7 growth stage. When damage was imposed at the V7 growth stage, no significant relationship between percent damaged plants and yield was observed ($F=0.97$; df 1, 55; $P=0.33$) with a mean removal of 23.8% of the above ground biomass (Figure 3.4).

An additional study was conducted to evaluate the sensitivity of vegetative growth stages to defoliation. Significantly more above ground biomass was removed at the V3 and V5 growth stages compared to growth stages V7 and V9 ($F=12.16$; df 3, 12; $P<0.01$) (Figure 3.5). Yield was significantly reduced when defoliation occurred during the V9 growth stage compared to the V5 growth stage and the non-damaged control ($F=2.92$; df 4, 57.73; $P=0.03$) (Figure 3.6).

Discussion

Manual removal of ca. 23% to 36% of the above ground biomass from 100% of plants during the V3 to V7 growth stages and fall armyworm infestations at the V7 growth stage did not impact yield. Loss of 17.6% of the above ground biomass from 100% of plants at the V9 growth stage reduced yield by ca. 20%. Based on these studies it does not appear that fall armyworm infestations occurring before the V8 growth stage are economically important for sorghum planted during the recommended window in Mississippi. These results are similar to Starks and Burton (1979) in that fall armyworm feeding during later sorghum growth stages may have significant influence on sorghum growth and yields. Further research is needed to determine the impact of greater levels of defoliation during the early vegetative growth stages. Also, the current study indicates that the sensitivity of sorghum to defoliation can increase at the V9 growth stage as significantly less defoliation resulted in lower yields at the V9 growth stage (just prior to boot stage) compared to more defoliation at the V3, V5, and V7 growth stages that did not impact yield. This indicates that treatment should be considered when infestations occur close to the beginning of the reproductive growth stages. Overall, little information is available on impact of defoliation during the vegetative growth stages on sorghum yield. Additional research is needed to determine the critical levels of infestation and defoliation to impact yield at specific growth stages.

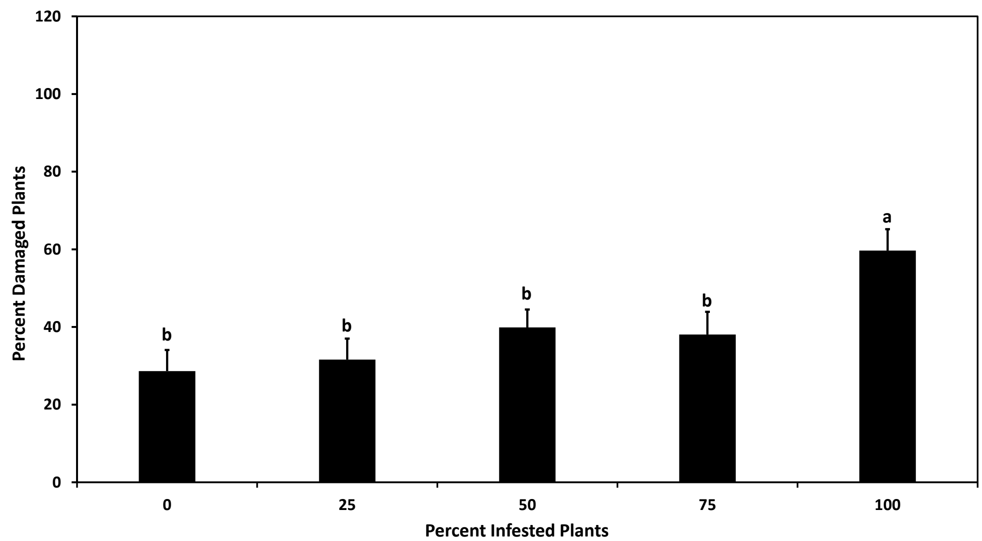

Figure 3.1 Mean (SE) percentage of grain sorghum, *Sorghum bicolor*, with fall armyworm, *Spodoptera frugiperda*, feeding damage from infestations conducted at the V7 growth stage in Stoneville and Starkville, MS in 2017. Means with a common letter are not different (Fisher's PLSD, P≤0.05).

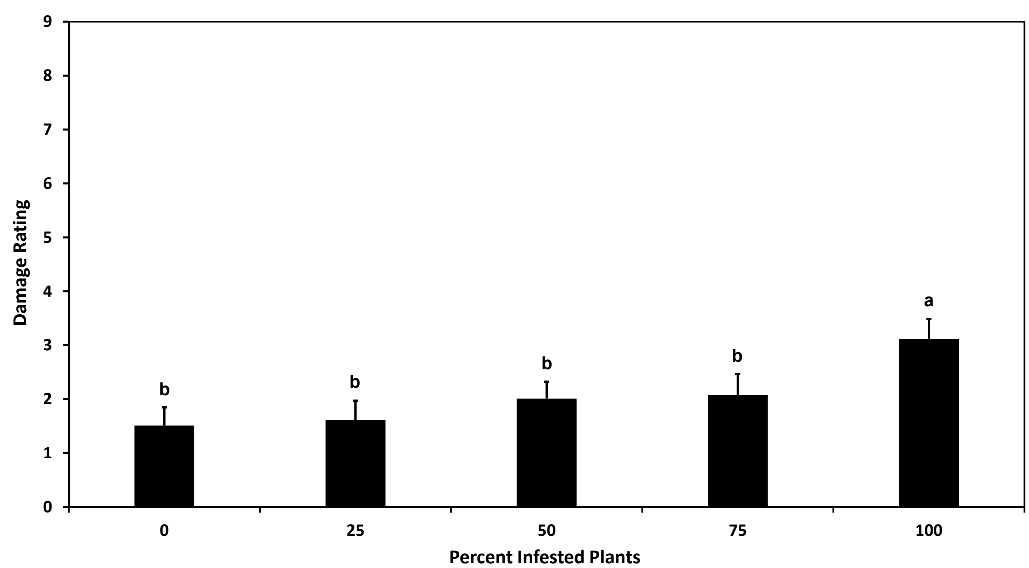

Figure 3.2 Mean (SE) damage rating per plot for artificial infestations to grain sorghum, *Sorghum bicolor*, with fall armyworm, *Spodoptera frugiperda*, conducted at the V7 growth stage in Stoneville and Starkville, MS in 2017. Bars with a common letter are not significantly different. (Fisher's PLSD, P≤0.05).

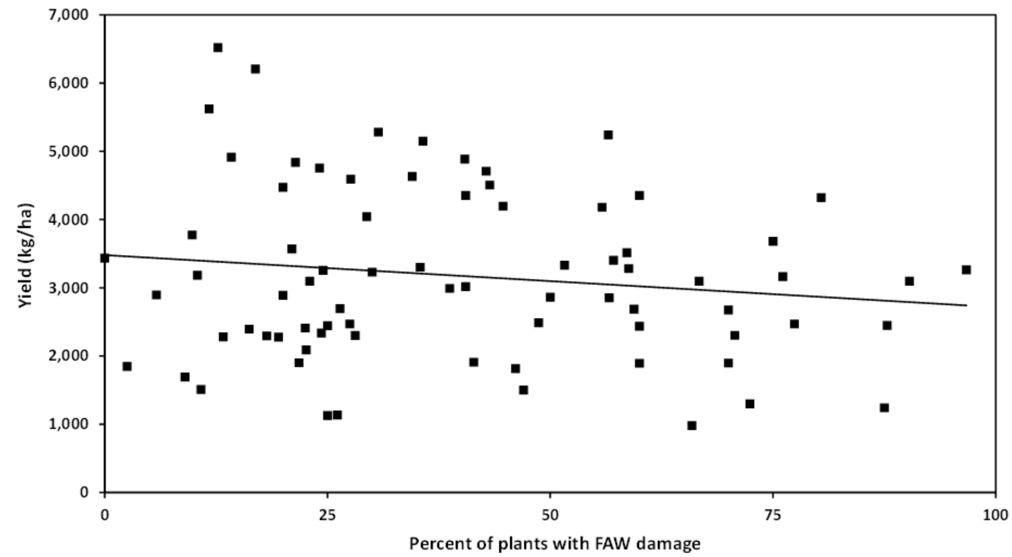

Figure 3.3 Relationship between percent of plants with fall armyworm, *Spodoptera frugiperda,* damage and grain sorghum, *Sorghum bicolor,* yield (P=0.23; kg/ha=3,522-6.3338x (±5.19)). Artificial infestations conducted at the V7 growth stage in Stoneville and Starkville, MS in 2017.

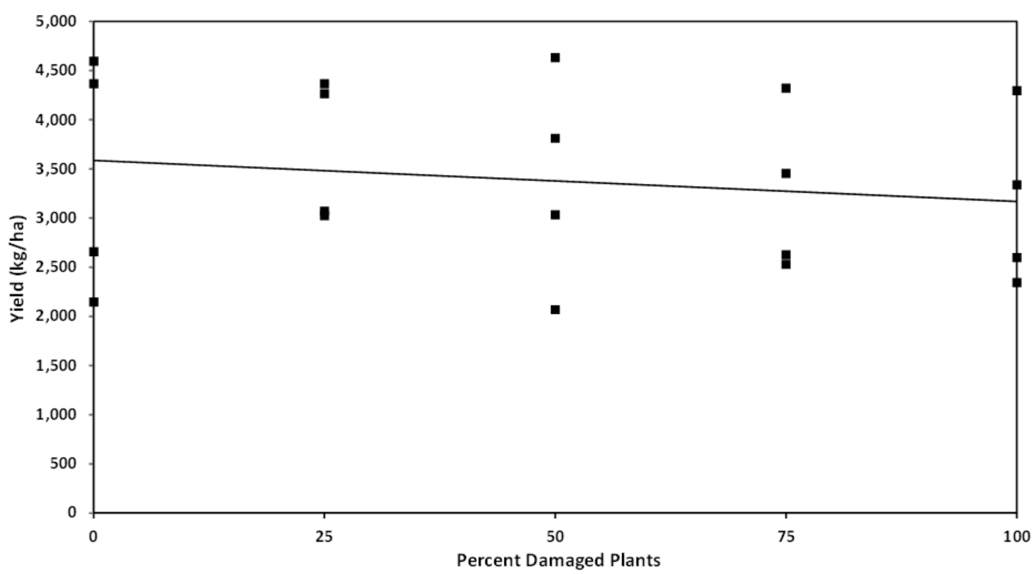

Figure 3.4 Relationship between percent damaged plants and grain sorghum, *Sorghum bicolor,* yield (P=0.33; kg/ha=3,562-3.7119x (±3.78)). Manual damage treatments imposed at the V7 growth stage in Stoneville and Starkville, MS in 2017.

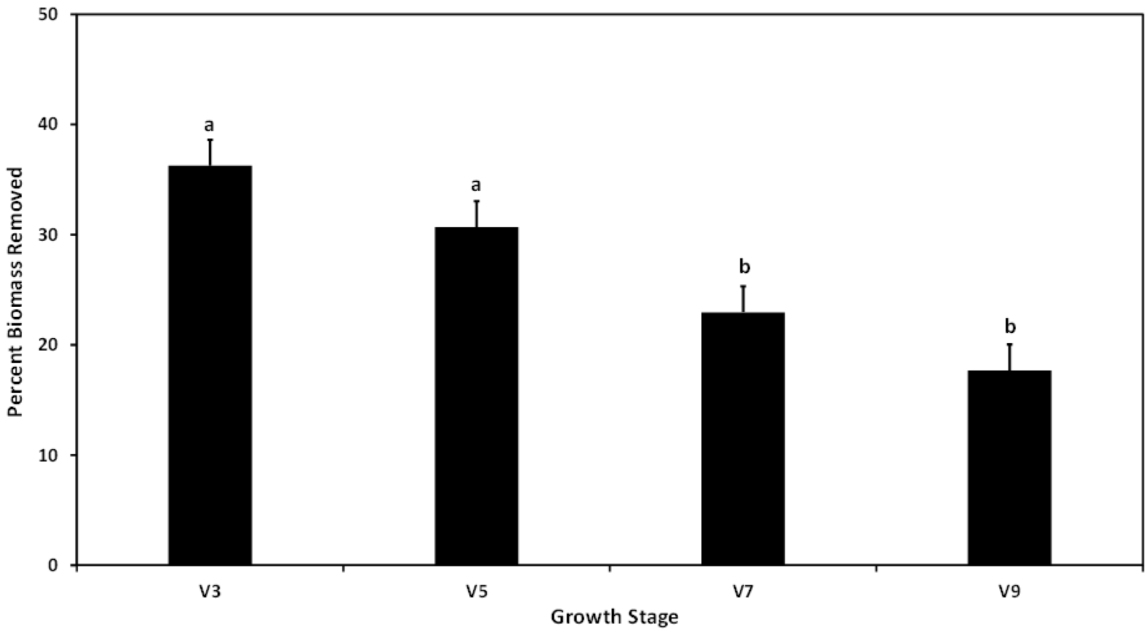

Figure 3.5 Percent of above ground biomass removed from grain sorghum, *Sorghum bicolor*, in Stoneville and Starkville, MS in 2017. Bars with a common letter are not significantly different (Fisher's PLSD, P≤0.05).

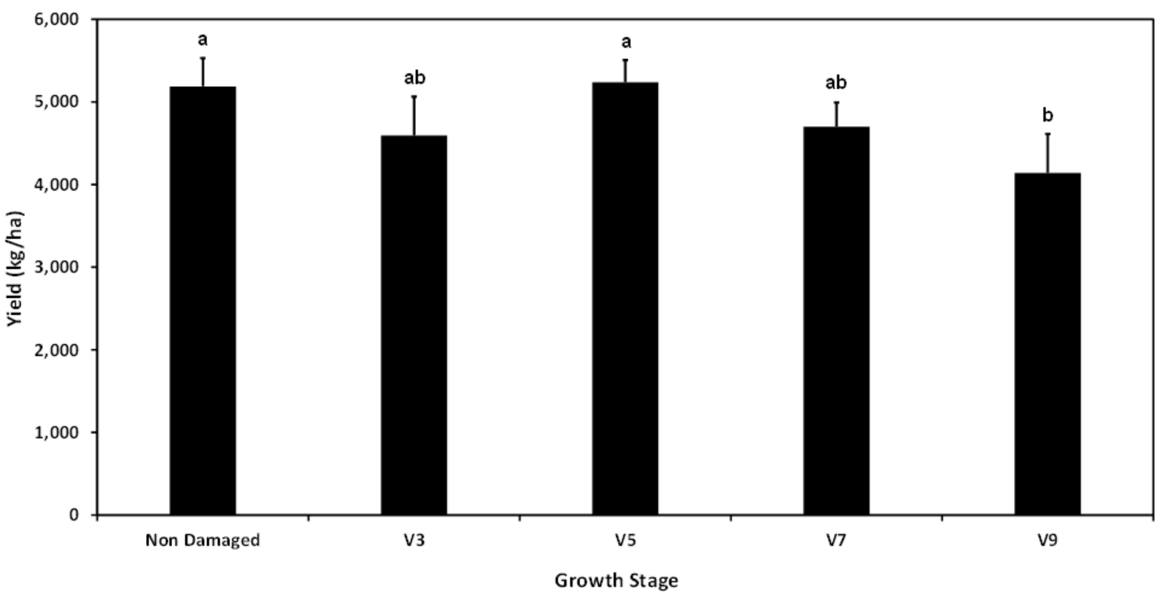

Figure 3.6 Impact of manual defoliation at selected vegetative growth stages on grain sorghum, *Sorghum bicolor*, yield in Stoneville and Starkville, MS in 2017. Bars with a common letter are not significantly different (P≤0.05, Fisher's PLSD).

Table 3.1 Damage scale used for damage assessment as described by Davis et al. (1992).

Score	Description
0	**No visible damage.**
1	**Only pinhole lesions present on whorl leaves.**
2	**Pinholes and small circular lesions present on whorl leaves.**
3	**Pinholes, small circular lesions and few elongated lesions of up to 1.3 cm in length present on whorl and/or furl leaves.**
4	**Small elongated lesions present on whorl leaves and a few mid-size elongated lesions of 1.3 to 2.5 cm in length present on whorl and/or furl leaves.**
5	**Small elongated lesions and several mid-size elongated lesions present on whorl and furl leaves.**
6	**Small and mid-size elongated lesions plus a few large elongated lesions of greater than 2.5 cm in length present on whorl and/or furl leaves.**
7	**Many small and mid-size elongated lesions present on whorl leaves plus several large elongated lesions present on the furl leaves.**
8	**Many small and mid-size elongated lesions present on whorl leaves plus many large elongated lesions present on the furl leaves.**
9	**Many elongated lesions of all sizes on whorl and furl leaves plus a few uniform to irregular shaped holes (basement membrane consumed) eaten from the base of the whorl and/or furl leaves.**

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